CALIFORNIA ENVIRONMENTAL PROTECTION AGENCY
DEPARTMENT OF PESTICIDE REGULATION
MEDICAL TOXICOLOGY BRANCH

SUMMARY OF TOXICOLOGY DATA
SULFOSULFURON

Chemical Code # 5136, Document Processing Number (DPN) # 52249
SB 950 # N/A
February 22, 2005

I. DATA GAP STATUS

Chronic toxicity, rat: No data gap, possible adverse effect

Chronic toxicity, dog: No data gap, no adverse effect

Oncogenicity, rat: No data gap, possible adverse effect

Oncogenicity, mouse: No data gap, possible adverse effect

Reproduction, rat: No data gap, no adverse effect

Teratology, rat: No data gap, no adverse effect

Teratology, rabbit: No data gap, no adverse effect

Gene mutation: No data gap, no adverse effect

Chromosome effects: No data gap, no adverse effect

DNA damage: No data gap, no adverse effect

Neurotoxicity: No data gap, no adverse effect

Toxicology one-liners are attached.

All record numbers for the above study types through 215576 (Document No. 52249-0040) were examined. This includes all relevant studies indexed by DPR as of 1/13/05.

NOTE: A U.S. EPA pesticide tolerance document has been prepared for Sulfosulfuron. This document stated that EPA had determined that mesenchymal tumors found in male mice in the 1997 Monsanto study represented a treatment response. The document is found at the following site: http://www.epa.gov/fedrgstr/EPA-PEST/1999/May/Day-19/p12247.htm

In the 1-liners below:
** indicates an acceptable study.

Bold face indicates a possible adverse effect.
indicates a study on file but not yet reviewed.

NOTE: This Summary of Toxicology Data makes extensive use of U.S. EPA executive summaries from their Data Evaluation Reports (DER's), after checking DER conclusions against the registrant's submitted reports for consistency. In each case, citations from EPA are preceded by the centered line:

"U.S. EPA Data Evaluation Report citation and executive summary (name of primary EPA reviewer):"

The EPA citation which follows is then left- and right- indented.

File name: t20050222.wpd
Original summary by C. Aldous, 2/22/05.

II. TOXICOLOGY ONE-LINERS AND CONCLUSIONS

These pages contain summaries only. Individual worksheets may identify additional effects.

COMBINED, RAT

****52249-0025 213508** Naylor, M. W. and F. A. Ruecker, "Combined chronic toxicity / oncogenicity study of MON 37500 administered in the diet to Sprague-Dawley rats," Monsanto Company, Environmental Health Laboratory (EHL), St. Louis, MO, 3/14/97. Monsanto Study No. ML-94-118, Laboratory Project No. EHL 94051. U.S. EPA MRID 44295759. See the U.S. EPA DER by L. Hansen, which tabulates all notable findings (including findings in the 20000 ppm males, which group was terminated on day 259 due to excessive toxicity, without routine inclusion in report summary data). The cited DER (which is found in DPR Document No. 52249-0040, Record No. 212856) is thorough, and data presented in the DER are consistent with the study report. All definitive findings from the study are recognized as such in the DER, and the conclusions by Dr. Hansen regarding meaningfulness of marginally elevated incidences were well justified. The DER may be used in lieu of a full DPR worksheet. Low incidences of urinary bladder adenoma (1/50) and urinary bladder carcinoma (1/50) in 5000 ppm females were considered by investigators and by the U.S. EPA reviewer to be treatment-related. Investigators considered these findings to meet the flagging criteria for an increase in uncommon tumors (p. 4 of report). Study is **acceptable**, with a **"possible adverse effect"** at dose levels which elicit urinary tract calculi. Aldous, 2/18/05. (See also DPR and/or EPA summaries of related studies, below).

U.S. EPA Data Evaluation Report citation and executive summary (reviewed by L. Hansen):

Naylor, M. W. and Ruecker, F. A. (1997) Combined Chronic Toxicity/Oncogenicity Study of MON 37500 Administered in the Diet to Sprague-Dawley Rats. Monsanto Company Environmental Health Laboratory (St. Louis, MO). Monsanto Study No. ML-94-118. Laboratory Project Number EHL 94051, March 14, 1997. MRID 44295759. Unpublished study.

SPONSOR: CEREGEN, Monsanto Company, St. Louis, MO

EXECUTIVE SUMMARY: In a combined carcinogenicity/chronic toxicity study (MRID 44295759), MON 37500 (tech., 98.4% a.i.) was administered to 60 Sprague-Dawley rats/sex/dose in the diet at dose levels of 0, 50, 500, 5,000 ppm and to 60 females at 20,000 ppm for 22 months. Surviving males at 20,000 ppm were sacrificed on day 259 due to excessive mortality (individual animal data only were provided for these males). Ten rats/sex/dose were sacrificed at 12 months. Average daily intake of test material was 0, 2.4, 24.4 or 244.2 mg/kg/day (males up to 5,000 ppm; 1,178.3 mg/kg/day, males at 20,000 ppm until day 259) and 3.1, 30.4, 314.1 or 1,296.5 mg/kg/day (females).

At 5,000 ppm, increased incidence/severity of abnormal crystals in the urine (both sexes, 70-100% affected vs. 10-40%, controls); slight, possibly treatment-related decrease in serum albumin in males (-20%); gross lesions of the urinary tract in females (calculi of kidney/bladder, dilatation of renal pelvis, 5 to 8.3% vs. 0-1.7%, controls) were observed. Microscopic findings in males included renal/urinary bladder calculi (3.3% and 5% vs. 0%, controls); dilatation of the renal pelvis (20% vs. 14%, controls); urinary bladder mucosal epithelial hyperplasia and dilatation (5% and 15% vs. 0% and 11%, controls); and increased incidence of mineralization in the heart, lung, pancreas and skeletal muscle (3.3% to 17% vs. 0 to 10%). In females, kidney pelvic epithelial hyperplasia (28% vs. 13%, controls), pelvic dilatation (13% vs. 0%), renal calculus (1.7% vs. 0%) and gastric pyloric erosions (10% vs. 1.7%) were observed. Females at interim sacrifice showed a slightly increased incidence of urinary tract calculi and related lesions. At 20,000 ppm, the following were observed: decreased survival in males (-33%, day 259) and possibly in females (-16%, termination); increased occurrence of blood-colored urine in males (37%, beginning on day 51) and intra-abdominal swelling (25%, day 135); decreased body weight/weight gain (-9%/-12% by sacrifice in males, up to -12%/-19%, days 51 to 485 in females); possible increased BUN at all time points in females (+20% to +60%); emaciated appearance in females (22% vs. 5%, controls); gross and microscopic lesions of the urinary tract in both sexes, including kidney or bladder calculi, dilatation, mineralization of renal cortex/medulla, bladder mucosal epithelial hyperplasia, pyelonephritis (females), squamous metaplasia of renal pelvic epithelium (females), increased severity of nephropathy (females), renal suppurative inflammation (females), necrosis of renal papilla (males) and hemorrhage of renal pelvis or bladder (males). In females, parathyroid hyperplasia (9% vs. 0%, controls), fibrous osteodystrophy of the femur and sternum (12% vs. 0%), and mineralization the aorta, cornea, heart, lungs, mesentery, pancreas, skeletal muscle, thyroid and spleen (3.7 to 18% vs. 0 to 1.7%, controls) were observed. **The LOEL is 5,000 ppm (244.2 mg/kg/day), based on increased incidence of urinary tract**

gross/microscopic lesions, mineralization in several tissues (males), abnormal urine crystals and possibly decreased albumin (males, termination). The NOEL is 500 ppm (24.4 mg/kg/day).

At 5,000 ppm, urinary bladder transitional cell carcinoma and papilloma were observed in females (1/50 each, vs. 0/50, controls) and were considered treatment-related because these tumors occur rarely. Dosing was adequate for both sexes based on increased urinary tract lesions and abnormal crystals in the urine at 5,000 ppm. Dosing at 20,000 ppm in males resulted in excessive toxicity. The HED Cancer Assessment Review Committee will evaluate the relationship of these tumors to treatment.

This study in the rat is classified **Acceptable** and satisfies the guideline requirement for a carcinogenicity/chronic toxicity study (83-5) in rodents.

52249-0026 213509 Naylor, M. W. [addendum to 52249-0025 213508, above], Dec. 2, 1997. This addendum, provided at the request of EPA reviewer (L. Hansen), shows historical control data for several kidney and urinary bladder lesions in Sprague-Dawley rats, obtained from Charles River Laboratories at Portage MI and tested in 16 chronic or oncogenicity studies at EHL from 1982 through apparently the time of this supplementary report. Terminology varied between pathologists and over time during this period. As a result, up to 5 equivalent descriptors were given for each type of lesion. Incidences of tabulated lesions in kidneys were: "calculus/microcalculus" [123/872 in F and 26/866 in M], hydronephrosis or pelvic dilatation [66/872 in F and 59/866 in M], pelvic epithelial hyperplasia [178/872 in F and 117/866 in M], and glomerulonephropathy [530/872 in F and 780/866 in M]. Urinary bladder lesions were "polyp/papilloma" or "#B - papilloma" [1/814 in F and 0/808 in M], "M - transitional cell carcinoma [0/814 in F and 0/808 in M], and epithelial hyperplasia [27/814 in F and 58/808 in M]. The relatively uncommon control incidences of neoplasia justify classifying the primary study as indicative of an oncogenic effect. Aldous, 2/18/05. No worksheet.

52249-0027 213510 Dubelman, S. [addendum to 52249-0025 213508, above], March 2000. This study evaluated the percentage of sulfosulfuron in bladder calculi samples from 3 randomly selected rats from the combined study [M #3049, 5000 ppm; F #4017, 20000 ppm, and F #4035, 20000 ppm]. Calculi samples (typically several mg in weight) were weighed, mashed with a spatula, and dissolved with sonication in acetonitrile/water, then assayed by HPLC with spectrophotometric detection against analytical sulfosulfuron standards. Two independent calculi samples were evaluated per rat, with two sub-samples each at different dilutions. Percent sulfosulfuron in rat calculi were 94% and 102% for the two 20000 ppm rats, and 71% for the 5000 ppm rat. No peaks other than for sulfosulfuron were evident in chromatograms shown for any of the rats. Thus it may be concluded that the calculi were entirely or almost entirely composed of sulfosulfuron. Aldous, 11/29/04. No worksheet.

52249-0029 213513 Cohen, S. M. and L. L. Arnold, "Effects of dietary sulfosulfuron on urinary parameters and the bladder in male Sprague-Dawley rats," Dept. of Pathology and Microbiology, Univ. of Nebraska Medical Center, Omaha, NE, March 5, 2001. Report No. 080100. Ten CRL:CD®(SD)IGS BR male rats/group were dosed for 10 weeks with 0, 50, 500,

5000, or 20000 ppm sulfosulfuron (97.4%) in diet. An additional group of 10 males was dosed with 5000 ppm sulfosulfuron plus 12,300 ppm NH_4Cl for the same duration. The addition of NH_4Cl acidifies the urine and reduces the extent of formation some crystals, such as magnesium ammonium phosphate [also called struvite: $Mg(NH_4)(PO_4)\cdot 6H_2O$, designated as $MgNH_4PO_4$ in the report]. Additional groups of ten rats were dosed with 0 or 20000 ppm sulfosulfuron for 10 weeks, then administered untreated diet for 10 weeks to evaluate recovery. Primary parameters assessed, based on experience of long term studies, were urinalysis, urinary crystal morphology, gross and light microscopic pathology of the urinary bladder, and scanning electron microscopy (SEM) of the bladder lumenal surface. The light microscopic examination included immunohistochemical evaluation of cellular proliferation in bladder urothelium following BrdU injection 1 hr prior to sacrifice. This is a supplementary study, not designed to place a NOEL. Nevertheless 5000 to 20000 ppm sulfosulfuron caused dose-related formation of "small round crystals" (consistent with sulfosulfuron aggregates), and was associated with decreased $MgNH_4PO_4$ crystal formation. Thus 500 ppm sulfosulfuron appears to be a "NOEL" for this study. Lack of bladder pathology in the absence of calculi (even in the presence of numerous crystals presumed to be sulfosulfuron) is a key finding in this study. Presence of papillary and nodular hyperplasia of the bladder epithelium and markedly elevated cellular proliferation index in (and only in) the single individual with bladder calculi suggests that such findings would only be obtained in association with calculi. Since, in retrospect, the only dose which produced definitive pathology was 20000 ppm, the choice to apply NH_4Cl supplementation only to a lower dose group had reduced utility. Also, particular attention to possible lesions of submucosal mesenchymal tissues (see mouse oncogenicity study) found no such changes at any dose level. Useful supplementary data, not relevant to FIFRA-mandated data requirements. Aldous, 2/18/05.

CHRONIC TOXICITY, RAT

(see combined, rat, above)

CHRONIC TOXICITY, DOG

**52249-0020 213503 Naylor, M. W. and F. A. Ruecker, "One year study of MON 37500 administered by capsule to beagle dogs," Monsanto Company, Environmental Health Laboratory (EHL), St. Louis, MO, and Experimental Pathology Laboratories (EPL), Herndon, VA, Feb. 5, 1997. Monsanto Study No. ML-95-199, Laboratory Project No. EHL 95042. This study was reviewed by L. Hansen at U.S. EPA under MRID 44295754. A copy of the EPA DER is found in DPR Document No. 52249-0040, Record No. 212853. A registrant's summary of the study is also found in DPR Document No. 52249-0040, Record No. 212844 (immediately following subchronic rat DER in that volume). DPR reviewer (Aldous) concurs with all key conclusions of the EPA review, as summarized in the DER executive summary. Study is acceptable, with no adverse effects. Aldous, 2/18/05.

U.S. EPA Data Evaluation Report citation and executive summary (reviewed by L. Hansen):

Naylor, M. W. and Ruecker, F.A. (1997) One Year Study of MON 37500 Administered by Capsule to Beagle Dogs. Monsanto Company Environmental Health Laboratory (St. Louis, MO). Monsanto Study No. ML-95-199, Laboratory Project No. EHL 95042 MSL 15007. February 5, 1997. MRID 44295754. Unpublished study.

SPONSOR: CEREGEN, Monsanto Company, St. Louis, MO

EXECUTIVE SUMMARY: In a chronic toxicity study (MRID 44295754) MON 37500 (tech., 98.5% a.i.) was administered to 5 beagle dogs/sex/dose by gelatin capsule at dose levels of 0, 5, 20, 100 or 500 mg/kg/day, 5 days/week) for 1 year. Controls received empty gelatin capsules.

At 500 mg/kg/day, abnormal urinary crystals were reported at 6 months in 1 male, which also developed urinary tract calculi and hemorrhage and thickened/irregular mucosa of the urinary bladder. Two males (including the above male) had yellow crystals in the urine on a total of 5 occasions in cageside observations. There were no compound related effects on mortality, clinical signs, body weight, food consumption, hematology, clinical chemistry, ophthalmology, or organ weights at any dose level. No treatment-related effects were observed in females. **The LOEL is 500 mg/kg/day, based on the presence of abnormal urinary crystals and bladder pathology secondary to formation of urinary tract calculi in males. The NOEL is 100 mg/kg/day.**

This chronic toxicity study is acceptable and satisfies the guideline requirement for a chronic oral study (83-1(b)) in the dog.

52249-0021 213504 Addendum to study 52249-0020 213503, above. At the request of the EPA reviewer, Monsanto provided an explanation by the study director (Naylor) of the procedures for recording clinical signs observed in between detailed weekly examinations. In addition, this record contains three pages of manually-recorded observations, including 5 notations of "yellow precipitate in urine," involving two high dose males. This information was included in the DER. There is no DPR worksheet for this record. Aldous, 10/28/04.

ONCOGENICITY, RAT

(see combined, rat, above)

ONCOGENICITY, MOUSE

****52249-0022 213505** Naylor, M. W. and D. C. Thake, "Oncogenicity study of MON 37500 administered in diet to CD-1® mice for 18 months," Monsanto Company, Environmental Health Laboratory (EHL), St. Louis, MO, and Experimental Pathology Laboratories (EPL), Herndon, VA, 2/19/97. Monsanto Study No. ML-94-119, Laboratory Project Nos. EHL 94052, MSL 15013. This study was reviewed by P. V. Shah at U.S. EPA under MRID 44295755. A copy of the EPA DER is found in DPR Document No. 52249-0040, Record No. 212854. DPR reviewer (Aldous) concurs with all key conclusions of the EPA review, as summarized in the DER executive summary. Study is acceptable, with possible adverse effects (based on benign urinary bladder mesenchymal tumors in the presence of associated non-neoplastic changes in the bladder). It should be noted that kidney tubular adenomas were observed in 2 high dose mice, vs. none in other groups. Examination of control and high dose kidney slides by renal pathologist, G. C. Hard, (reported in DPR Record No. 213507, and discussed in the DPR review for Record No. 213505) found no treatment-associated increases in predisposing lesions such as atypical tubular hyperplasia, suggesting that the tubular adenomas were most likely incidental. Aldous, 2/18/05.

U.S. EPA Data Evaluation Report citation and executive summary (reviewed by P. V. Shah):

M. W. Naylor and D. C. Thake,1997, Oncogenicity Study of MON 37500 Administered in Diet to CD-1 Mice for 18 Months, Monsanto Company (CEREGEN), Environmental Health Laboratory, 645 S. Newstead, St. Louis, MO and Experimental Pathology Laboratories, Inc.(EPL), P. O. Box 474, Herndon, VA, Monsanto Study No. ML-94-119; Laboratory Project No. EHL 94052, MSL 15013, RD 1353, February 19, 1997. MRID 44295755. Unpublished Study.

SPONSOR: Monsanto Agricultural Company

EXECUTIVE SUMMARY: In a carcinogenicity study (MRID 44295755), MON 37500 (98.4% a.i.) was continuously administered to 60 CD-1 albino mice/sex/dose as a dietary admixture at dose levels of 0, 30, 700, 3000 or 7000 ppm (0, 4.0, 93.4, 393.6 or 943.5 mg/kg/day to males or 0, 6.5, 153.0, 634.9 and 1388.2 mg/kg/day to females) for 18 months. Ten animals/sex/dose were sacrificed at month 12 for hematology, limited serum chemistry analyses and gross/microscopic pathology.

At 3,000 ppm, increased incidence of calculus in the urinary bladder (34% vs. 5%, controls) and microscopic lesions of the bladder caused by calculus formation (mucosal epithelial hyperplasia, 42% vs. 6.8%, controls; chronic inflammation, 39% vs. 3.4%; and ulceration, 5.1% vs. 0%) were reported in males. At 7,000 ppm in males, clinical signs of toxicity (urine-stained fur, intra-abdominal swelling and abnormal penile erection; 6-12 affected animals vs. 1-2, controls), dilatation of the renal pelvis (45% vs. 8.3%), dilatation of the bladder (13.3% vs. 0%) were also observed. There were no treatment-related effects on survival, body weight, food consumption or hematology parameters in males and

no treatment-related effects were reported in females at any dose tested. **The LOEL is 3,000 ppm (393.6 mg/kg/day), based on gross and microscopic effects related to urinary calculus formation in the urinary bladder of males. The NOEL is 700 ppm (93.4 mg/kg/day).**

At 7,000 ppm, increased incidence of benign mesenchymal tumors of the urinary bladder in males (5/50 or 10% vs. 0%, controls; not statistically significant) was observed. One male at 3,000 ppm also had this tumor (and 1 control and 1 high dose female). Single incidences of renal tubular adenoma were observed in 1 male and 1 female at 7,000 ppm. The HED Cancer Assessment Review Committee will evaluate the relationship of these tumors to treatment. Dosing in males was considered adequate based on urinary tract effects. No toxicity was reported in females, but dosing was considered adequate because the high dose exceeded 1,000 mg/kg/day.

This carcinogenicity study in the CD-1 mice is **acceptable and does satisfy** the guideline requirement for a carcinogenicity study (83-2*(b)*) in mice.

52249-0024 213507 Hard, G. C., "Expert report on renal histopathologic changes in a mouse study with MON 37500 (Sulfosulfuron): Addendum to oncogenicity study of MON 37500 administered in diet to CD-1 mice for 18 months," (4 Aug. 1999), Monsanto Study No. ML-94-119, MRID No. 44295755. Dr. Hard, a renal specialist, examined kidney slides from all control mice and all high dose mice in the oncogenicity study (Record No. 213505). He confirmed the presence of the two tumors identified by the original study pathologist. He sought for evidence of predisposing lesions at the high dose, such as atypical tubular hyperplasia, but found no pathological response which would be expected in chemically-induced oncogenic responses in kidney. These observations, associated with descriptions of progressive stages of pre-neoplastic renal lesions and of chronic progressive nephropathy, are included in the review of the DPR oncogenicity study review. This report was noted in the worksheet for DPR Record No. 213505. Aldous, 11/30/04.

52249-0023 213506 Naylor, M. W. Addendum to: "Oncogenicity study of MON 37500 administered in diet to CD-1® mice for 18 months" (DPR Record No. 213505). This additional information was provided at the request of the U.S. EPA reviewer, P. V. Shah, on the subject of tumor observations in the cited study. Historical control data from 16 studies reaching equivalence criteria showed kidney tubular adenoma incidences of 0/953 for males, and 4/956 for females. The comparative rarity of this tumor type suggests that even the one such tumor per sex at 7000 ppm should not be dismissed as a plausible treatment effect, despite the observation by Dr. Hard (see review of DPR Record No. 213505) that predisposing renal lesions were absent. Historical control data from 16 studies on benign mesenchymal urinary bladder tumors showed incidences of 1/910 for males and 0/931 for females. Investigators and U.S. EPA had previously stipulated an oncogenic effect in 7000 ppm males, and causality in the case of the single 3000 ppm male with this tumor type cannot be excluded. The present record includes reproductions of three published articles on mouse urinary bladder tumors of mesenchymal origin. The first two articles provide case histories on small numbers of tumors, with detailed lesion descriptions and some keys to differential diagnoses. The Halliwell article draws from the

earlier two submissions as well as additional observations from a broad base of studies including about 100 submucosal mesenchymal tumors or "SMT's" evaluated by an ILSI Risk Science Institute working group. That article noted that these tumors (1) tend to remain localized within the submucosal area, (2) usually are found in the (mesodermally-derived) trigone area of the bladder, and are commonly too small to be identified at necropsy (hence reported incidences of these tumors, which are often small, may be largely influenced by sectioning technique), (3) SMT's have usually been reported in CD-1 or Swiss Webster mice, and only at very low incidence in other strains of mice, (4) older literature used various designations for these tumors, such as leiomyosarcoma or hemangiopericytoma, (5) some of SMT's have been associated with crystalluria, (6) morphologically similar tumors sometimes arise in the seminal vesicles [also of mesodermal origin, these being derived from the mesonephros], (7) consistent with presumed mesodermal origin, immunohistochemical evaluations of these tumors are negative or inconclusive for collagen, cytokeratin, or striated muscle, but positive for desmin (associated with smooth muscle), (8) nevertheless the tumors do not typically have the appearance of smooth muscle neoplasms, and (9) physical obstruction leading to bladder distention may be a frequent or major factor in SMT development. Aldous, Dec. 1, 2004 (no worksheet).

Published articles included in Document No. 52249-0023

Jacobs, J. B. *et al.*, "Chemically induced smooth muscle tumors of the mouse urinary bladder," Cancer Research 36, 2396-2398 (1976).

Chandra, M. and C. H. Frith, "Spontaneously occurring leiomyosarcomas of the mouse urinary bladder," Toxicologic Pathology 19, 164-167 (1991).

Halliwell, W. H., "Submucosal mesenchymal tumors of the mouse urinary bladder," Toxicologic Pathology 26, 128-136 (1998).

REPRODUCTION, RAT

**52249-0019 213502 Naylor, M. W., and D. C. Thake, "Two generation reproduction study of MON 37500 administered in the diet to albino rats," Monsanto Company, Environmental Health Laboratory (EHL), St. Louis, MO, Jan. 10, 1996. Monsanto Study No. ML-94-120, Laboratory Project No. EHL 94053. This study was reviewed by L. Hansen at U.S. EPA under MRID 44295758. A copy of the EPA DER is found in DPR Document No. 52249-0040, Record No. 212852. Groups of 30 Sprague-Dawley rats/sex/group were dosed in diet with Sulfosulfuron in a guideline 2-generation reproduction study. This technical is designated as MON 37500 in report, and is 98.4% purity). Doses were 0, 50, 500, 5000, or 20000 ppm. DPR reviewer (Aldous) concurs with all key conclusions of the EPA review. Study is acceptable, with no adverse effects. Aldous, 2/18/05. There is a brief discussion in the DPR worksheet.

U.S. EPA Data Evaluation Report citation and executive summary (reviewed by L. Hansen):

Naylor, M. W. and Thake, D. C. (1996) Two Generation Reproduction Study of MON 37500 Administered in the Diet to Albino Rats. Ceregen, Monsanto Company Environmental Health Laboratory (St. Louis, MO). Laboratory study number ML-94-120, Project No. EHL 94053. January 10, 1996. MRID 44295758. Unpublished.

SPONSOR: Monsanto Company, St. Louis, MO

EXECUTIVE SUMMARY: In a 2-generation reproduction study (MRID 44295758), MON 37500 (tech., 98.4% a.i.) was administered to 30 Sprague-Dawley rats/sex/dose/generation in the diet at dose levels of 0, 50, 500, 5,000 or 20,000 ppm (during premating, equivalent to average daily intake for P adults of 0, 3.1, 31.6, 312.1 and 1312.8 mg/kg/day, males and 0, 3.6, 36.2, 363.2 and 1454.1, females; for F1a adults, 0, 3.1, 31.1, 315.8 and 1,378.8, males and 0, 3.7, 37.7, 377.8 or 1,598.0 mg/kg/day, females).

At 20,000 ppm, statistically significant decreases in mean cumulative body weight gain during premating in P females (-12.2% less than controls), mean body weight of F1a males through premating week 8 and females throughout premating (both ~ -15%, week 0, decreasing to ~ -6% to -9% by week 11 due to lower week 0 weights; gains not significantly lower), and mean body weight/ weight gain during gestation in both generations (-8%/-16%, P and -10%/-6%, F1a) were observed. Body weights were significantly lower during lactation (P ~ -8 to -9%, days 0 and 7; F1a ~ -7% to -10%, days 0-21) due to reduced weight during gestation, not decreased weight gain. Slight decreases in food consumption were observed in P females during gestation days 0-7 and 7-14 (about -10%). Increased incidence of several urinary tract gross/microscopic lesions (including calculi, hydronephrosis, renal pelvic epithelial hyperplasia, nephropathy) were observed, mostly in P and F1a females. Deaths of 2 P and 1 F1a males appeared to be treatment-related and were associated with calculus formation and urinary tract pathology. Slightly increased relative kidney weight in P females, F1a males and F1a females (ranging from 7% to 17%) may have been treatment-related. No treatment-related reproductive toxicity, clinical signs of toxicity, organ weight changes, pre-weaning pup weight changes or pup gross findings were observed. **The reproductive toxicity NOEL is ≥20,000 ppm (1,312.8 mg/kg/day) and the LOEL is >20,000 ppm. The parental systemic toxicity LOEL is 20,000 ppm based on decreased parental body weight and/or weight gain during premating, gestation and lactation, mortality (males) and increased incidence of urinary tract pathology related to calculus formation. The parental systemic toxicity NOEL is 5,000 ppm (312.1 mg/kg/day). The offspring toxicity LOEL is 20,000 ppm (1,312.8 mg/kg/day) based on decreased body weight gain in post-weaning adolescent rats, and the offspring NOEL is 5,000 ppm (312.1 mg/kg/day).**

This reproduction study in the rat is classified Acceptable and satisfies the guideline requirement for a 2-generation reproductive study (OPPTS 870.3800, §83-4) in the rodent.

TERATOLOGY, RAT

**52249-0016 213498 Holson, J. F., "A developmental study of MON 37500 in rats," WIL Research Laboratories, Inc., May 10, 1994. Monsanto Study No. WI-93-261, Laboratory Project No. WIL-50226. This study was reviewed by L. D. Chitlik at U.S. EPA under MRID 44295756. A copy of the EPA DER is found in DPR Document No. 52249-0040, Record No. 212848. Groups of 25 mated CRL:CD®BR females were dosed on gestation days 6-15 by gavage with Sulfosulfuron in a guideline developmental toxicity study. This technical is designated as MON 37500 in the report. Purity was 99.1%. Vehicle was corn oil (5 ml/kg b.w.). Doses were 0, 100, 300, or 1000 mg/kg/day. There was no maternal nor developmental toxicity evident at any tested dose level, the high dose being the limit test. Maternal and developmental toxicity NOEL's are 1000 mg/kg/day. DPR reviewer (Aldous) concurs with all key conclusions of the EPA review. Study is acceptable, with no adverse effects. Aldous, 10/19/04.

U.S. EPA Data Evaluation Report citation and executive summary (reviewed by L. D. Chitlik):

Holson, J. F.,et al, (1994) A Developmental Study of MON 37500 in Rats. WIL Research Laboratories, Inc., Ashland, Ohio 44805-9281 for Monsanto Agricultural Company. May 10, 1994. WIL- 50226. MRID 44295756. Unpublished

EXECUTIVE SUMMARY:

In a developmental toxicity study (MRID44295756) MON 37500 Technical (99.1% purity, Lot No. NPD-9209-4523-T) was administered in Mazola corn oil to 25 Sprague-Dawley Crl:CD BR female rats by gavage at dose levels of 0, 100, 300, and 1000 mg/kg/day from days 6 through 15 of gestation.

No overt maternal toxicity was observed at any dose level in this study. However, since the highest dose level in this study was equivalent to the limit dose (1000 mg/kg/day), the dosing is considered adequate to meet regulatory requirements and additional testing at higher levels is not required. The **NOEL for maternal toxicity is greater than 1000 mg/kg/day (HDT). A LOEL was not determined.**

No dose related increases in developmental toxicity are apparent at any dose level in this study. **Therefore, the NOEL for developmental toxicity is greater that 1000 mg/kg/day (HDT). A LOEL was not determined in this study.**

Classification: This study is considered **acceptable/guideline** and fulfills the regulatory requirement for a rat developmental toxicity study.

TERATOLOGY, RABBIT

**52249-0017 213499 Holson, J. F., "A developmental study of MON 37500 in rabbits," WIL Research Laboratories, Inc., 11/21/94. Monsanto Study No. WI-94-002, Laboratory Project No. WIL-50227. This study was reviewed by L. D. Chitlik at U.S. EPA under MRID 44295757. A copy of the EPA DER is found in DPR Document No. 52249-0040, Record No. 212849. Groups of 20 mated NZW females were dosed on gestation days 7-19 by gavage with Sulfosulfuron in a guideline developmental toxicity study. This technical is designated as MON 37500 in report, and is >98.5% purity). Vehicle was methylcellulose (2 ml/kg b.w.). Doses were 0, 50, 250, or 1000 mg/kg/day. There was no maternal nor developmental toxicity evident at any dose level, the high dose being the limit test. Maternal and developmental toxicity NOEL's are 1000 mg/kg/day. DPR reviewer (Aldous) concurs with all key conclusions of the EPA review. Study is acceptable, with no adverse effects. Aldous, 10/21/04.

U.S. EPA Data Evaluation Report citation and executive summary (reviewed by L. D. Chitlik):

> Holson, J. F., et al, (1994) A Developmental Toxicity Study of MON 37500 in Rabbits. WIL Research Laboratories, Inc., Ashland, Ohio 44805-9281 for Monsanto Agricultural Company. WIL-50227. MRID# 44295757. Unpublished
>
> EXECUTIVE SUMMARY:
>
> In a developmental toxicity study (MRID# 44295757) MON 37500 Technical (98.5% purity, Lot No. GHQ-9307-5385-T) was administered as a mixture in 0.5% methylcellulose to 20 New Zealand white female rabbits by gavage at dose levels of 0, 50, 250, and 1000 mg/kg/day from days 7 through 19 of gestation.
>
> No overt maternal toxicity was observed at any dose level in this study. However, since the highest dose level in this study was equivalent to the limit dose (1000 mg/kg/day), the dosing is considered adequate to meet regulatory requirements and additional testing at higher levels is not required. The **NOEL for maternal toxicity is greater than 1000 mg/kg/day (HDT). No LOEL for maternal toxicity was determined in this study.**
>
> No dose related increase in developmental toxicity is apparent at any dose in this study. **Therefore, the NOEL for developmental toxicity is greater than 1000 mg/kg/day (HDT). No LOEL for developmental toxicity was observed in this study.**
>
> Classification: This study is classified as **acceptable/guidelines** and fulfills the regulatory requirement for a rabbit developmental toxicity study.

NOTE: DPR Document No. 52249-0040, Record No. 212850 is a continuation of the DER cited under Record No. 212849.

52249-0018 213501 Historical control data relevant to 52249-0017 213499, above. The EPA reviewers had requested control data by study with completion dates, particularly with respect to two defects with nominal increases in treated rabbits ("extra site of ossification anterior to sternebra #1," and "sternebrae with thread-line attachment"). These historical control data for 66 studies were presented sequentially, over the period of 1982 through 1997 for these two lesion types. Incidences in treated groups in the sulfosulfuron study were well within historical range for contemporary studies. This information was considered in the DER cited above. This volume also presents historical data by study for numerous other malformations and variations observed in external, skeletal, and visceral examinations. Aldous, 10/19/04 (not relevant information for a DPR worksheet).

GENE MUTATION

**52249-0030 213514 Stegeman, S. D., J. Warren, and L. D. Kier, "Ames/Salmonella mutagenicity assay of MON 37500," Monsanto Study No. ML-94-004, 3/15/95. This study was reviewed by I. Mauer at U.S. EPA under MRID 44295760. A copy of the EPA DER is found in DPR Document No. 52249-0040, Record No. 212857. The DER justifiably considers this study to be acceptable, with no adverse effects. The associated executive summary is suitable for DPR use. Aldous, 12/29/04.

U.S. EPA Data Evaluation Report citation and executive summary (reviewed by I. Mauer):

> Stegeman, S.D., Warren, J. and Kier, L. D. (1995). Ames/Salmonella Mutagenicity Assay of MON 37500, performed at Monsanto's Environmental Health Laboratory (EHL), St. Louis MO, Lab. Project No. EHL 94002/Monsanto Study No. ML-94-004, Final Report dated March 15, 1995. MRID 44295760.
>
> SPONSOR: Monsanto Company, T. Louis MO
>
> EXECUTIVE SUMMARY: In a microbial reverse gene mutation study (MRID No. 44295760), Salmonella typhimurium strains TA1535, TA1537, TA98, TA100 and TA102 were exposed to MON 37500 (>98.5%) doses ranging from 5-5000 µg/plate +/-S9 using both the standard plate incorporation assay and the preincubation modification to the standard assay. The S9 fraction was derived from Aroclor 1254-induced Sprague Dawley male rat livers and MON 37500 was delivered to the test system in dimethyl sulfoxide.
>
> Cytotoxicity was seen for the majority of strains at doses ≥1500 µg/plate +/-S9 using both testing procedures. All strains responded in the expected manner to the appropriate positive control. **There was, however, no evidence that MON 37500 induced a mutagenic effect in any strain at any noncytotoxic dose using either procedure.**

This study is classified as Acceptable and satisfies the guideline requirements for a bacterial gene mutation assay (84-2).

**52249-0031 213515 Stegeman, S. D., L. D. Kier, K. L. Asbury, S. L. Garrett, and J. G. McAdams, "CHO/HGPRT gene mutation assay of MON 37500," Monsanto Study No. ML-95-200, Jan. 5, 1996. This study was reviewed by I. Mauer at U.S. EPA under MRID 44295761. A copy of the EPA DER is found in DPR Document No. 52249-0040, Record No. 212858. The DER justifiably considers this study to be acceptable, with no adverse effects. The associated executive summary is suitable for DPR use. Aldous, 12/30/04.

U.S. EPA Data Evaluation Report citation and executive summary (reviewed by I. Mauer):

Stegeman, S.D., Kier, L. D., Asbury, K. L., Garrett, S.L. and McAdams, J. G. (1996). CHO/HGPRT Gene Mutation Assay of MON 37500; Monsanto Co. Environmental Health Laboratory, St. Louis, MO; Study No. ML-95-200; Study Completion Date: January 5, 1996. (Unpublished) MRID NUMBER: 44295761

SPONSOR: Monsanto Co., St. Louis, MO

EXECUTIVE SUMMARY: In a mammalian cell gene mutation assay (MRID No. 44295761), cultures of normal ($HGPRT^+$) Chinese hamster ovary (CHO) cells were exposed for 3 hours to doses of 624-5000 µg/mL MON 37500 (98.5%) in the absence and presence of 1, 5 or 10% S9 (initial trial). The confirmatory trial investigated five doses ranging from 312-5000 µg/mL -/+5% S9. The S9 homogenate was derived from Aroclor 1254-induced rat livers and the test material was delivered to the test system in Ham's F12 medium (without serum).

MON 37500 was insoluble at ≥2500 µg/mL. Marginal cytotoxicity (≈40% reduction in cell survival) was seen at 5000 µg/mL without S9. The S9-activated test material and lower nonactivated concentrations (≤2500 µg/mL) were not cytotoxic. The positive controls induced the expected mutagenic responses. **There was, however, no evidence that MON 37500 was mutagenic at any dose under any assay condition.**

This study is classified as Acceptable and satisfies the guideline requirement for a gene mutation assay (§84-2).

CHROMOSOME EFFECTS

**52249-0032 213516 Murli, H., "Chromosomal aberration study in human whole blood lymphocytes with a confirmatory assay with multiple harvests with MON 37500," Monsanto Study No. HL-95-201, March 11, 1996. This study was reviewed by I. Mauer at U.S. EPA under MRID 44295762. A copy of the EPA DER is found in DPR Document No. 52249-0040, Record No. 212859. The DER justifiably considers this study to be acceptable, with no adverse effects. The associated executive summary is suitable for DPR use. There is an error in the Table 3 of

the DER, corresponding to Table 4 of the report: the DER table heading indicates a 19.5 hr treatment, whereas the source table states that treatment was 41.4 hr. Aldous, 2/22/05.

U.S. EPA Data Evaluation Report citation and executive summary (reviewed by I. Mauer):

Murli, H. (1996) Chromosome Aberration Study in Human Whole Blood Lymphocytes with a Confirmatory Assay with Multiple Harvests with MON 37500. Corning Hazleton Inc. (CHV), Laboratory Project No. 17146-0-449CO, Final Report dated March 11, 1996. (Unpublished) MRID NUMBER: 44295762.

SPONSOR: Monsanto Co., St. Louis, MO 63110

EXECUTIVE SUMMARY: In an in vitro cytogenetic assay (MRID No. 44295762), cultured human lymphocytes, obtained from a single donor, were exposed to MON 37500 in initial (3-hr exposure ±S9 to MON 37500, followed by 17-hr incubation and then 2-hr exposure to colcemid) and confirmatory (-S9: 19.5- and 43.4-hr exposures to MON 37500, followed by colcemid; +S9: same as initial) assays. The S9 homogenate was derived from Aroclor 1254-induced rat livers and the test material was delivered to the test system as a solution in DMSO.

The highest dose level evaluated (±S9) in all assays and harvest times was 1000 µg/mL. A precipitate was observed at this dose level. In addition, reductions (from 14.9 to 66.7%) were seen in mitotic indices at this dose level ±S9. However, there were no indications of any increased incidence of cells with chromosomal aberrations and/or numbers of polyploid cells. The positive controls induced the expected high yields of metaphase spreads with chromosomal aberrations. **Based on the these considerations, it is concluded that MON 37500 did not induce a clastogenic response in human lymphocytes under the conditions of this assay either in the presence or absence of S9 at doses up to and including those associated with cytotoxicity and/or test material precipitation..**

This study is classified as ACCEPTABLE satisfying the requirement for FIFRA Test Guideline (84-2) for mammalian in vitro chromosome aberration data.

52249-001 155496 "Chromosomal aberration test of TKM-19 using cultured mammalian cells." (Yoshida, N., Hita Research Laboratories, for Takeda Chemical Industries, Ltd., September, 1996). U.S. EPA MRID 44280201. TKM-19 (sulfosulfuron, 98.9%, white crystalline powder) was tested with Chinese hamster lung fibroblasts (CHL cells, clone no. 11) without and with S9 mix (from Sprague-Dawley male rats induced with phenobarbital/5,6-benzoflavone). Cells were exposed without S9 for 24 hours to 0 (0.5% carboxymethyl cellulose, 10% of culture medium), 1000, 2000 or 3000 µg/ml and for 48 hours to 0, 2000, 2500 and 3000 µg/ml, duplicate cultures per concentration. In addition, cells were exposed for 6 hours with and without activation at 0, 1250, 2500 or 5000 µg/ml in duplicate followed by 18 hours incubation.

Positive controls were mitomycin C without activation and cyclophosphamide with activation. Two slides were prepared per culture with 100 cells scored per culture for chromatid- and chromosome-type aberrations and for numerical aberrations. All concentrations were above the limit of solubility of sulfosulfuron, with precipitation. Growth was inhibited to a considerable extent in the 24 and 48 hour cultures at 2000 μg/ml and above. The results were judged to be positive for chromosomal aberrations without S9 at 24 hours (2000 and 3000 μg/ml) and at 48 hours (2000, 2500 and 3000 μg/ml) but only marginal at 6 hours without activation at 5000 μg/ml, the other cultures being negative after the 6-hour exposure. The % of cells with aberrations (minus gaps) increased with dose and with time of exposure at the same concentration. The positive controls were functional. There was no evidence of an induction of polyploidy. **Possible adverse effect** (an increase in chromosomal aberrations with and without activation at concentrations resulting in precipitation). Interpretation of the results is complicated by the precipitation at all concentrations. Hence, the report is **unacceptable**. (J. Gee, July 8, 1998).

NOTE: A U.S. EPA review (contracted to Oak Ridge National Laboratory, then signed by I. Mauer at HED) also noted increased chromosomal aberrations, but only above the level of solubility in non-activation conditions. That DER (found in DPR Document No. 52249-0040, Record No. 212861) classified this study as "acceptable/guideline." See below.

U.S. EPA DER citation and executive summary (reviewed by I. Mauer):

Anonymous (1996) Chromosomal aberration test of TKM-19 using cultured mammalian cells. Hita Research Laboratories, Chemical Biotesting Center, Chemicals Inspection & Testing Institute, Japan, 822, 3-chome, Ishii-machi, Hita, Oita 877, Japan. Laboratory report number T-4500, September 17, 1996. MRID 44280201. Unpublished

SPONSOR: Takeda Chemical Industries, LTD., 10, Wadai, Tsukuba, Ibaraki 300-42, Japan

EXECUTIVE SUMMARY: In a mammalian cell cytogenetics assay (MRID 44280201), CHL cell cultures were exposed to TKM-19 (98.9% a.i., Lot No. 30013919) in 0.5% sodium carboxymethylcellulose (CMC) at concentrations of 1000, 2000 or 3000 μg/mL for 24 hours without metabolic activation (S9-mix), at concentrations of 2000, 2500 or 3000 μg/mL for 48 hours without S9-mix and at concentrations of 1250, 2500 or 5000 μg/mL for 6 hours with and without S9-mix. The S9-fraction was obtained from phenobarbital/5,6-benzoflavone induced male Sprague-Dawley rat liver.

TKM-19 was tested up to cytotoxic and precipitating concentrations. Two preliminary cytotoxicity tests were performed. The first test used DMSO as the solvent but the maximum achievable concentration, 2500 μg/mL, was not sufficiently cytotoxic. In the second preliminary cytotoxicity test, 0.5% CMC was used as the solvent and concentrations up to 5000 μg/mL were achieved although a precipitate was seen in the culture medium at concentrations of 1000 μg/mL and higher. TKM-19 reduced cell growth in a dose-dependent manner to

14.1% of the solvent control after 24 hours treatment without S9-mix and to 6.1% of the solvent control after 48 hours treatment without S9-mix. Metaphase cells were rare or absent at 4000 and 5000 µg/mL at 24 and 48 hours. A six-hour treatment reduced cell growth to 46.9% of the solvent control at 5000 µg/mL without S9-mix but to only 78.3% with S9-mix. In the cytogenetic assay, 200 cells/dose/ treatment time/activation condition were scored for structural and numerical chromosomal aberrations. A dose-related increase in the percentage of cells with structural chromosomal aberrations (virtually all chromatid breaks and exchanges) was seen, including or excluding gaps, in the 24 hour and 48 hour treatment cultures (without S9-mix). The percentage of cells with structural chromosomal aberrations, excluding gaps, reached 19.5% at 3000 µg/mL in the 24 hour treatment cultures (2% in the solvent control) and 85.5% at 3000 µg/mL in the 48 hour treatment cultures (1% in the solvent control). Positive control values were 58% and 65.5% at 24 and 48 hours, respectively. Results from the six hour treatment without S9-mix met the testing laboratory's criterion for a "suspect" positive at 5000 µg/mL, reaching 6.5% including gaps; however, gaps are not generally included in chromosomal aberration evaluation and when they were excluded the percentage of cells with aberrations was 3.5%, a negative response. Results from the six hour treatment with S9-mix were negative at all doses, including or excluding gaps. No increases in numerical aberrations were seen in the study. Positive and solvent control values were appropriate throughout the study. **There was evidence of structural chromosomal aberration induction over background but only under non-activated conditions, and at concentrations exceeding the limit of solubility of the test article.**

There is no consensus in the research community on the suitability of doses beyond the solubility limits of the test material or on the relevance of results obtained at those doses (see the Report of the International Workshop on Standardization of Genotoxicity Test Procedures, Mutation Research 312(3), 1994). The FIFRA Test Guidelines state that relatively insoluble substances should be tested up to the limit of solubility. They also state that the highest dose tested should show evidence of cytotoxicity or reduced mitotic activity. To reach the desired cytotoxicity level, all doses in the present study exceeded the solubility limits of the test material. This study, therefore, is classified as **acceptable/guideline**. It satisfies the requirement for FIFRA Test Guideline 84-2 for in vitro cytogenetic mutagenicity data.

DNA DAMAGE

**52249-0033 213517 Stegeman, S. D., L. D. Kier, S. L. Garrett, J. G. McAdams,, J. Warren, and S. Schermes, "Mouse bone marrow micronucleus assay of MON 37500," Monsanto Study No. ML-94-005, March 1, 1995. This study was reviewed by I. Mauer at U.S. EPA under MRID 44295763. A copy of the EPA DER is found in DPR Document No. 52249-0040, Record No. 212863. The DER justifiably considers this study to be acceptable, with no adverse effects. U.S. EPA justifiably agreed with the study investigators that the study was negative, despite a statistically significant increase in micronuclei/1000 PCE's among mid-dose females at 24 hr

harvest time only, for reasons adequately clear in the DER and in the original report. The associated executive summary is suitable for DPR use. Aldous, 12/31/04.

U.S. EPA Data Evaluation Report citation and executive summary (reviewed by I. Mauer):

Stegeman, L. D.; Kier, L. D.; Garrett, S. L.; McAdams, J. G.; Warren, J. and Schermes, S. (1995) Mouse Bone Marrow Micronucleus Assay of MON 37500, performed at Monsanto's Environmental Health Laboratory (EHL), Lab Project No. EHL 94003/Monsanto Study No. ML-94-005, Final Report dated March 1, 1995 (Unpublished) MRID NUMBER: 44295763.

SPONSOR: Monsanto Co., St. Louis, MO 63110

EXECUTIVE SUMMARY: In an in vivo mammalian cytogenetics (micronucleus) assay (MRID No. 44295763), groups of CD-1® mice (5M:5F/dose/sacrifice time) were gavaged orally with single doses of solvent carrier (Tricaprylin, 10 mL/kg) or MON 37500 (1250, 2500 and 5000 mg/kg), and sacrificed 24, 48 and 72 hrs post-dosing. A positive control group of 5M and 5F received a single oral dose (40 mg/kg) of cyclophosphamide and was sacrificed 24 hrs later. Slides were prepared from harvested bone marrow, and were evaluated for presence of micronucleated polychromatic erythrocytes (MPCE's) as well as possible cytotoxicity (ratio of PCEs to total erythrocytes).

No mortalities occurred during the micronucleus assay; hypoactivity was observed in one animal in the 2500 mg/kg group and one at 5000 mg/kg. There was no evidence of target cell cytotoxicity (a significant decrease in the PCE/total erythrocyte ratio). The positive control (cyclophosphamide, 40 mg/kg, administered as a single oral gavage dose in tricaprylin) induced the expected high yield of MPCE's in mice sacrificed at 24 hrs. The mean incidence of MPCE's was statistically ($p \leq 0.01$) elevated in 2500 mg/kg females sacrificed at 24 hours, but the value (2.7/1000 PCEs; concurrent vehicle controls: 1.0/1000 PCEs) was within historical corn oil vehicle range (0-7.4/1000 PCEs), and the incidence in 5000 mg/kg females sacrificed at 24 hours was not significantly elevated (1.4/1000 PCEs), indicating the statistically significant elevation at 2500 mg/kg was a sporadic event. **MON 37500 (>98.5%) did not induce a clastogenic effect in either sex at any sacrifice time.**

In addition, the Agency has received a mouse bone marrow assay of ^{14}C-radiolabeled MON 37500 (in MRID 44295764) which demonstrates that MON 37500 (and/or its metabolites) was present in the bone marrow of male CD-1^{R} mice at 2 and 8 hours following oral gavage dosing of 2000 mg/kg MON 37500 (administered in corn oil, at 10 mL/kg).

The study, with its negative findings, is classified as Acceptable and satisfies the requirements for FIFRA Test Guideline 84-2 for a micronucleus assay.

52249-0036 213520 Stegeman, S. D., L. D. Kier, L. A. Albin, and H. Lau, "Mouse bone marrow assay of ^{14}C radiolabeled MON 37500," Monsanto Study No. ML-95-289, April 8, 1996. This supplementary study was reviewed by I. Mauer at U.S. EPA under MRID 44295764. A copy of the EPA DER is found in DPR Document No. 52249-0040, Record No. 215573. This study was conducted to confirm that orally-dosed sulfosulfuron reached the marrow site in sufficient amounts for a valid micronucleus test (supporting study 52249-0033 213517, above). The DER's executive summary is suitable for DPR use. Aldous, 12/31/04.

U.S. EPA Data Evaluation Report citation and executive summary (reviewed by I. Mauer):

Stegeman, L. D.; Kier, L. D.; Albin, L. A. and Lau, H. (1996) Mouse Bone Marrow Assay of ^{14}C Radiolabeled MON 37500, performed at Monsanto's Environmental Health Laboratory (EHL), Lab Project No. EHL 95106/Monsanto Study No. ML-95-289, Final Report dated April 8, 1996 (Unpublished) MRID NUMBER: 44295764.

SPONSOR: Monsanto Co., St. Louis, MO 63110

EXECUTIVE SUMMARY: In a pharmacokinetic study in mice (MRID 44295764), four CD-1 males were administered ^{14}C-labeled MON 37500 (Sulfosulfuron) in corn oil (approximately 10 µCi/mouse) at 2000 mg/kg. Two animals were sacrificed at 2 hours post-dose, and two at 8 hours post-dose. Two males received corn oil only; one was sacrificed 2 hours later and the other 8 hours later. Blood and femoral bone marrow were processed and radioactivity was measured by liquid scintillation counting (LSC).

Significant amounts of radioactivity were detected in blood plasma and bone marrow at both sacrifice times from dosed animals (2 hours: blood plasma: 1292 µg equivalents/g tissue; bone marrow: 415 µg equivalents/g tissue; 8 hours: blood plasma: 324 µg equivalents/g of tissue; 29 µg equivalents/g of tissue). These findings demonstrate transport of parent compound and/or its metabolites to bone marrow, and indicates that target cell exposure occurred in the mouse micronucleus assay (MRID 44295763).
This ancillary study is ACCEPTABLE (non-guidelines), although it does not meet or satisfy any specific guideline requirement (including FIFRA Test Guideline §85-1). It was designed to demonstrate that MON 37500 and/or its metabolites reach mouse bone marrow following administration via oral gavage, as supporting data to a mouse micronucleus assay (in MRID 44295763).

NEUROTOXICITY

52249-0034 213518 Branch, D. K., D. C. Thake, T. A. Kaempfe, and A. A. Li, "Acute neurotoxicity study of MON 37500 in Sprague-Dawley rats," Monsanto Study No. ML-95-286, 1/30/97. No noteworthy findings were observed, and no further information is required of this study. This study was reviewed by L. Hansen at U.S. EPA under MRID 44295749. That U.S. EPA DER is found in DPR Document No. 52249-0040, Record No. 212865. DPR reviewer

(Aldous) concurs with all key conclusions of the EPA review. It should be noted that although that review's executive summary describes the dose levels in units of "mg/kg/day," it is clear from context that this was indeed an acute study in which each animal received a single oral dose. Acceptable with no adverse effects. Aldous, Jan. 12, 2005 (no worksheet).

U.S. EPA Data Evaluation Report citation and executive summary (reviewed by L. Hansen):

> Branch, D. K., Thake, T. A., Kaempfe T. A. and Li, A. A. (1997) Acute Neurotoxicity Study of MON 37500 in Sprague-Dawley Rats. CEREGEN, Monsanto Company Environmental Health Laboratory (St. Louis, MO). Study No. ML-95-286; Laboratory project number EHL 95100; MSL 14959, January 30, 1997. MRID 44295749. Unpublished study.
>
> SPONSOR: Monsanto Company, St. Louis, MO
>
> EXECUTIVE SUMMARY: In an acute oral neurotoxicity screening study (MRID 44295749), MON 37500 (tech., 98.5% a.i.) was administered to 10 Sprague-Dawley rats/sex/dose by gavage in corn oil (8 ml/kg body weight) at dose levels of 0, 125, 500 or 2000 mg/kg/day (actual dose levels were approximately 20% higher for all groups, based on analytical values for dosing solutions). Functional observational battery and motor activity testing were conducted pretest, at time of peak effect (7 hrs post-dosing) and at 7 and 14 days post-dosing. Neurohistopathological examinations were performed on 5 perfused animals/sex of the control and high dose groups.
>
> No treatment-related effects on clinical signs, body weight, food consumption, functional observational battery parameters, motor activity, gross pathology or neuropathology were observed. **The NOEL is ≥2,000 mg/kg/day. The LOEL is >2,000 mg/kg/day.**
>
> This acute neurobehavioral screening study is classified **Acceptable** and satisfies the guideline requirement (81-8SS) in the rat.

**52249-0035 213519 Kaempfe, T. A., D. C. Thake, D. K. Branch, J. A. Warneke, F. L. Speck, and A. A. Li, "Subchronic neurotoxicity study of MON 37500 administered in feed to Sprague-Dawley® rats." Monsanto Study No. ML-96-045, 1/20/97. This study was reviewed by L. Hansen at U.S. EPA under MRID 44295753. That U.S. EPA DER is found in DPR Document No. 52249-0040, Record No. 212866. DPR reviewer (Aldous) concurs with all key conclusions of the EPA review. It should be noted that the DER considered the 20000 ppm dose in males to be a systemic LOEL, based on slight (not statistically significant) body weight decrements. Somewhat stronger evidence of a body weight effect in 20000 ppm males was noted by EPA reviewers to be found in the subchronic rat study (MRID 44295750, found in DPR Document No. 52249-0040, Record No. 212843, beginning on p. 96 of that volume). Neurotoxicity NOEL ≥ 20000 ppm. Acceptable, with no adverse effects (no worksheet). Aldous, 2/22/05.

U.S. EPA Data Evaluation Report citation and executive summary (reviewed by L. Hansen):

Kaempfe T. A., Thake, D. C., Branch, D. K., Warneke, J. A., Speck, F. L. and Li, A. A. (1997) Subchronic Neurotoxicity Study of MON 37500 in Sprague-Dawley Rats. CEREGEN, Monsanto Company Environmental Health Laboratory (St. Louis, MO). Study No. ML-96-045; Laboratory project number EHL 96012; MSL 14915, January 20, 1997. MRID 44295753. Unpublished study.

SPONSOR: Monsanto Company, St. Louis, MO

EXECUTIVE SUMMARY: In a subchronic oral neurotoxicity screening study (MRID 44295753), MON 37500 (tech., 98.5% a.i.) was administered to 10 Sprague-Dawley rats/sex/dose in the diet at levels of 0, 200, 2,000 or 20,000 ppm (corresponding to average daily doses of 0, 12, 122 or 1,211 mg/kg/day in males and 0, 14, 141 or 1,467 mg/kg/day in females). Functional observational battery and motor activity testing were conducted pretest and at weeks 4, 8, and 13. Neurohistopathological evaluations were performed on 5 perfused animals/sex from control and high dose groups.

At 20,000 ppm, body weight/weight gain showed marginal but non-statistically significant decreases in males (gradually increasing to -6%/-11% at 13 weeks). This decrease is considered a threshold effect because significant reductions were observed at 20,000 ppm in the rat 90-day oral toxicity study (MRID 44295750). No treatment-related effects on clinical signs, food consumption, functional observational battery parameters, motor activity, gross pathology or neuropathology were observed. **The study NOEL (threshold) is 20,000 ppm (1,211 mg/kg/day), based on marginal reductions in body weight/weight gain of males. The LOEL is >20,000 ppm (>1,211 mg/kg/day).**
This subchronic neurotoxicity screening study is classified **Acceptable** and satisfies the guideline requirement (82-7SS) in the rat.

RAT METABOLISM

**52249-0037 213521 Lau H. et al., "The absorption, distribution, elimination and metabolism of MON 37500 in Sprague-Dawley rats following oral and intravenous administration," Monsanto Study No. MSL 14300. See the U.S. EPA DER by J. Peggins in DPR Document No. 52249-0040, Record No. 215576, under MRID 44295765. The executive summary presented in the DER had no serious discrepancies with the original study report. Acceptable, with no adverse effects. Aldous, 2/22/05.

U.S. EPA Data Evaluation Report citation and executive summary (reviewed by J. Peggins):

H. Lau, M.L. Kinnett, L. A. Albin, J. Warren, R. C. Chott, H. Fujiwara, R. G. Nadeau and M. E. Holland, The Absorption, Distribution, Elimination and Metabolism of MON 37500 in Sprague-Dawley Rats Following Oral and Intravenous Administration. (1996), Environmental Health Laboratory.

Laboratory report number 93041, Monsanto Study# 14300, January 5, 1996. MRID 44295765. Unpublished.

SPONSOR: Monsanto Agricultural Co.

EXECUTIVE SUMMARY: In a metabolism study (MRID 44295765), radiolabelled MON 37500 (98% radiochemically pure a.i.), was administered to Sprague-Dawley rats. In the pilot study, 2-3 rats/sex/group were administered either ^{14}C-Pd MON 37500 (radiolabel at the C-5 position of pyrimidine ring) or ^{14}C-Im MON 37500 (label at the C-3 position of the imidazopyridine ring), each at 10 mg/kg or 1,000 mg/kg, by gavage. Rats dosed with Im or Pd MON 37500 isotopes had similar metabolite profiles; therefore in the main study, 5 rats/sex/group were administered a mixture of ^{14}C-Pd, ^{14}C-Im, ^{13}C-Im and ^{12}C-MON 37500 at single oral doses of 10 or 1,000 mg/kg, repeated oral doses of 10 mg/kg (14 consecutive daily unlabelled doses, followed by 1 radiolabelled dose), or a single intravenous dose of 10 mg/kg.

More than 90% of the administered radioactivity was excreted by 72 hrs post-dosing (mean total recovery ranged from 97% to 101%). Between 77% to 87% of the administered radioactivity was excreted in the urine in all low dose groups (5-13% was excreted in the feces, low single and repeat oral dosing and low IV dosing), whereas feces was the major route of elimination at high dose (63%, males and 55%, females; urinary excretion 32%, males and 33%, females). Biliary excretion appeared to occur, as shown by fecal excretion in the intravenous dose group (9% and 5% of administered dose, males and females). Urinary excretion followed a biexponential pattern with half-lives of 2.2-5.8 hrs, initial phase, and 21.4-56.7 hrs, terminal phase (similar rates were observed for whole body elimination). In all dose groups, minimal radioactivity was retained in tissues (less than 0.07%) or carcass (less than 0.17%). In the pilot study, expiration of $^{14}CO_2$ was insignificant (<0.04% of administered dose, all groups) and therefore not measured in the main study. Absorption was essentially complete at low dose (95%, males and 91%, females) but markedly reduced at the high dose (36%, males and 39%, females); it was not determined whether this was a dose-dependent effect or was due to a dose-vehicle effect. Metabolism of MON 37500 in all groups was minimal and most was excreted unmetabolized (for example, it represented more than 90% of the total radioactivity excreted in the urine). MON 37500 was metabolized primarily via ring hydroxylation of the a-position carbon of the pyrimidine ring, or demethylation of the methoxy group at the 4- or 6-position of the pyrimidine ring. In addition, a limited amount of cleavage of the sulfonamide bond into sulfonamide or other imidazopyridine compounds and pyrimidine compounds was observed. Major metabolites identified in urine and feces were desmethyl MON 37500 (3.6% or greater of administered dose), 5-hydroxy MON 37500 (1.9% or greater) and sulfonamide (2.6% or greater). A fourth, Feces 488 (ND-1.68%; feces only), was partially characterized as a desmethylated metabolite. Other minor metabolites, 1 imidazopyridine and 3 pyrimidine metabolites, were also found at low levels (<1%) in the feces. Metabolite profiles were qualitatively similar among animals

given high vs. low doses, single vs. repeated doses, oral vs. intravenous doses, or between males and females (the percentages of different metabolites varied but due to the low levels present, variations were not considered biologically significant). It was noted that small amounts of white precipitates were observed in the urine of high dose rats at 12 and 24 hrs post-dosing. These were not characterized, but were probably unmetabolized MON 37500, based on the low level of biotransformation.

This metabolism study in the rat is classified acceptable and satisfies the guideline requirement for a metabolism study (§85-1) in rats.

SUBCHRONIC STUDIES

**52249-0012 213494 Naylor, M. W. and D. C. Thake, "Pilot three month study of MON 37500 administered in feed to Sprague-Dawley rats," Monsanto Company, Environmental Health Laboratory (EHL), St. Louis, MO, 2/22/95. Monsanto Study No. ML-93-63, Laboratory Project No. EHL 93002. See the U.S. EPA DER by L. Hansen under MRID 44295750, which tabulates all notable findings. All definitive findings from the study are recognized as such in the DER, and the conclusions by Dr. Hansen regarding meaningfulness of marginally elevated incidences were well justified. The DER justifiably considered the 20000 ppm group to have plausibly shown a marginal response in kidney and urinary bladder at the 20000 ppm level, considering what is known from the rat combined study. The investigators also considered the same dose level as the LOEL, but strictly on the basis of body weight decrements in high dose males, and also in high dose females during late gestation only. There were no other subchronic findings, and no indications of reproductive effects in this pilot study. See executive summary of the DER for methods and findings (the entire DER is found in DPR Document No. 52249-0040, Record No. 212843). Acceptable, with no adverse effects. Aldous, Jan. 3, 2005.

U.S. EPA Data Evaluation Report citation and executive summary (reviewed by L. Hansen):

M. W. Naylor and D. C. Thake (1995) Pilot Three Month Study of MON 37500 Administered in Feed to Sprague-Dawley Rats. Monsanto Company Environmental Health Laboratory (St. Louis, MO). Study No. ML-93-63; Laboratory Project No. EHL 93002. February 22, 1995. MRID 44295750. Unpublished study.

SPONSOR: Monsanto Chemical Company, St. Louis, MO

EXECUTIVE SUMMARY: In a subchronic toxicity study (MRID 44295750), MON 37500 (tech., 98.9% a.i.) was administered to 10 male and 20 female Sprague-Dawley rats/dose in the diet at dose levels of 0, 20, 200, 2,000, 6,000 or 20,000 ppm (equivalent to average daily intake of 0, 1.2, 12.1, 123.2, 370.3 or 1,277.5 mg/kg/day, males and 0, 1.5, 14.6, 144.3, 447.5 or 1,489.1 mg/kg/day, females). Ten of the females/dose were mated 1:1 at week 10 with males of the

same dose group for up to 7 days in a 1-generation range-finding study. Reproductive and litter parameters were evaluated through lactation day 4.

At 20,000 ppm, mean body weight gain in males was statistically significantly lower than controls at termination (-18%; -28% by comparing gain calculated as percent of initial body weight). Body weight was also lower (-9.4%; not significant). Gain in pregnant females was less than controls (-26%; significant negative trend). When 1 dam with a dead litter was excluded, gain was -15% (not analyzed statistically). In females, a possible increase in incidence of gross and microscopic renal lesions associated with renal calculi (calculus, dilated pelvis, pyelonephritis, hyperplasia of pelvic epithelium, necrotic calcified debris; 2/10 affected) and urinary bladder mucosal epithelial hyperplasia (1/10) was observed. One male had renal calculi and bladder hyperplasia. No treatment-related effects on clinical observations, non-pregnant female body weight, food consumption, ophthalmology, hematology, clinical chemistry parameters, or reproductive/litter parameters were observed. **The systemic toxicity LOEL is 20,000 ppm (1,277.5 mg/kg/day), based on decreased body weight/weight gain in males, possible decreased weight gain in pregnant females during gestation days 14-21, and possibly renal lesions related to formation of calculi. The NOEL is 6,000 ppm (370.3 mg/kg/day).**

This subchronic toxicity study is classified **Acceptable** and satisfies the guideline requirement for a subchronic oral study (82-1a) in the rat. (The reproductive component of this study is not classified because it was conducted solely as a range-finding study for a later 2-generation reproduction study.)

**52249-0013 213495 Naylor, M. W. and D. C. Thake, "Three month study of MON 37500 administered by capsule to beagle dogs," Monsanto Company, Environmental Health Laboratory (EHL), St. Louis, MO, Jan 8, 1996. U.S. EPA MRID 44295751. Monsanto Study No. ML-94-324, Laboratory Project No. EHL 94108. See the U.S. EPA DER by J. O. Peggins, which tabulates all notable findings. All definitive findings from the study are recognized as such in the DER. The most definitive effect defining the LOEL is the presence of unidentified urinary crystals in several males and females at 300 and 1000 mg/kg/day, for which tabulated data are found in the report and the DER. See executive summary of the DER for methods and findings (the entire DER is found in DPR Document No. 52249-0040, Record No. 212845). Acceptable, with no adverse effects. No DPR worksheet. Aldous, Jan. 5, 2005.

U.S. EPA Data Evaluation Report citation and executive summary (reviewed by J. O. Peggins):

Naylor, M. W. and Thake, D. C. Summary: Naylor, M. W., (1996). Three Month Study of MON 37500 Administered by Capsule to Beagle Dogs. CEREGEN, A Unit of Monsanto Company Environmental Health Laboratory (EHL) St. Louis, Missouri 63110 (U. S. A.)EHL report# 94108, Monsanto# ML-94-324. MRID 44295751, 8 January 1996, Unpublished.

SPONSOR: Monsanto Agriculture Co.

EXECUTIVE SUMMARY: In a subchronic toxicity study (MRID 44295751) MON 37500 (98.4% a.i.) was administered to 50 beagle dogs (5 Males/5 Females/dose) by gelatin capsule at dose levels of (0, 30, 100, 300 and 1,000 mg/kg/day)for 90 days. Controls received empty gelatin capsules.
MON 37500's toxic effects were expressed primarily in the urinary tract and appeared to be secondary to formation of urinary calculi at 300 and 1000 mg/kg/day. Abnormal crystals (unidentified) were observed in the urine at day 45 at 300 and 1,000 mg/kg/day (males, 1 and 2, respectively; females, 3 and 2, respectively) and in females at study termination (3 at 300 mg/kg/day and 4 at 1,000 mg/kg/day). Hemorrhage, ulceration, inflammation, and/or mucosal epithelial hyperplasia in the urinary bladder were observed in one female in the 1000 mg/kg/day dose group and one in the 300 mg/kg/day dose group. Two males at 1000 mg/kg/day also showed treatment-related effects. One high dose male was sacrificed due to advanced urolithiasis and associated complications throughout the urinary tract, which included glomerulonephritis, degeneration of renal tubular epithelium, suppurative inflammation of the renal pelvis, arteritis/periarteritis, congestion, tubular protein accumulation and fibrin deposition in the capsular/pericapsular areas. Hemorrhage, erosions and ulcerations with acute inflammation, and degeneration of the tunica muscularis were also observed in the urinary bladder. Inflammation, edema and hemorrhage of periureteral tissue were associated with ureter damage. Inflammation and epithelial necrosis occurred in the urethra. Hemorrhage, acute inflammation and vasculitis/perivasculitis were noted in the prostate gland; the relationship of these lesions to treatment was unclear. The investigators concluded that necrosis of thymic lymphocytes in this male was probably due to release of adrenal cortical hormones subsequent to stress. A second male dog had bladder lesions of acute inflammation, erosions/ulcerations and edema. There were no differences in clinical signs, body weight/weight gain, food consumption, ophthalmologic results or hematology and clinical chemistry parameters related to administration of the MON 37500.

The systemic toxicity LOEL is 300 mg/kg/day, based on lesions in the urinary bladder in females occurring subsequent to urinary crystal formation/urolithiasis and on abnormal urinary crystals in males and females. The NOEL for systemic toxicity is 100 mg/kg/day.

This subchronic toxicity study in dogs is classified acceptable and satisfies the guideline requirement for a subchronic oral study (82-1b) in Dogs.

MISCELLANEOUS

52249-0028 213512 Healy, C., "Studies on the relevance of kidney and urinary bladder tumors in chronic studies with sulfosulfuron," project is designated R. D. No. 1536, 03/21/2001. This is a response to a U.S. EPA classification of sulfosulfuron with respect to oncogenicity, hence may be of interest in DPR risk assessment. This is not an original source of laboratory animal data. No DPR worksheet. Aldous, 12/31/04.